261986H
TX-90-5

ACUTE TOXICITY OF
90% min. 4-METHYLCYCLOHEXANE METHANOL

SYNONYM: MCHM

HAEL NO. 89-0081 ACC. NO. 907670

BY KENNETH P. SHEPARD, B.S.

TOXICOLOGICAL SCIENCES LABORATORY
HEALTH AND ENVIRONMENT LABORATORIES
EASTMAN KODAK COMPANY
ROCHESTER, NY 14652-3615

DATE OF STUDY COMPLETION JANUARY 26, 1990

261986H
TX-90-5

SUMMARY OF ACUTE TOXICITY STUDIES

Chemical: 4-Methylcyclohexane methanol
Synonym: MCHM

Accession No.: 907670

HAEL No.: 89-0081

Source Reference I.D. No.: X20511-3-3

Source: Eastman Kodak Company

Date of First Study Initiation: July 24, 1989

Comments: All animals were identified by metal ear tags/cage number. All specimens, raw data, and the final report of this work are stored in the archives of the Health and Environment Laboratories. Only limited analyses have been completed on the strength, purity, composition, stability, uniformity, and concentration of the test material. Professionals involved in this study other than the study director included: Gordon J. Hankinson, D.V.M., M.S., Laboratory Animal Medicine and Milan S. Vlaovic, D.V.M., Ph.D., Pathologist. Deviations from approved protocols or standard operating procedures included: None.

Director, Mammalian Toxicology Section
Study Director

January 26, 1990
Date

Director, Toxicological Sciences Laboratory

January 26, 1990
Date

ACUTE ORAL TOXICITY IN THE RAT

Dose levels tested: 625, 1250, and 2500 mg/kg

No. Rats/dose: 5 of each sex/dose

Vehicle: None

Solution/suspension: Administered as received

Initial Body weight range (g): Males: 204 - 245 Females: 153 - 188

Strain: Crl:CD®(SD)BR

Method of calculation: Weil method

Test SOP No.: TA 110, TA 120

DOSE mg/kg	NO. RATS DOSED(M,F)	NO. DEATHS (M,F)	TIME OF DEATH	WEIGHT GAIN* 1 WEEK(M,F)	WEIGHT GAIN* 2 WEEK(M,F)
			----------	5+,5+	5+,5+
625	5,5	0,0		5+,--	5+,--
1250	5,5	0,5	Day 1 to Day 3	--,--	--,--
5000	5,5	5,5	Day 1		

* + = Number of animals gaining weight
- = Number of animals losing weight

SUMMARY OF CLINICAL SIGNS

DOSE (mg/kg)	CLINICAL SIGN	NO. OF RATS AFFECTED
625	Slight Weakness Progressing to Moderate Weakness	5M,5F
	Vasodilatation	5M,5F
	Brown Urine	1F
	Clinically Recovered	5M,5F
1250	Slight Weakness	5M
	Moderate Weakness	5M,5F
	Severe Weakness	5M,5F
	Vasodilatation	5M,5F
	Brown Urine	1F
	Decreased Feces	1F
	Hypothermia	1F
	Prostration	1F
	Clinically Recovered	5M
	Death	5F
2500	Severe Weakness	5M,5F
	Prostration	5M,5F
	Vasodilatation	5M,5F
	Death	5M,5F

ACUTE ORAL TOXICITY IN THE RAT CONT.

ESTIMATED ORAL LD_{50} IN RATS

MALES: 1768 mg/kg - 95% C.I. 1340 - 2333 mg/kg
FEMALES: 884 mg/kg - 95% C.I. 670 - 1166 mg/kg

REMARKS: The test article was slightly toxic by the oral route. For the selection of dose levels, a range finding study was conducted utilizing one animal of each sex per dose level, with body weights ranging between 154-214 grams. Dose levels of 5000, 2500, 1250, 625, and 312 mg/kg were used in the range finding study, with the test material administered as received. All animals given a dose of 5000 or 2500 mg/kg, and the female at the 1250 mg/kg dose level were found dead on the day following administration of the test material. No other deaths occurred over a 14-day observation period. Based on the range finding study, dose levels of 625, 1250, and 2500 mg/kg were chosen for the oral toxicity study.

In the acute oral toxicity study, abnormal clinical signs at a dose level of 2500 mg/kg included severe weakness, vasodilatation, and prostration in all animals on the day of administration of the test material. All females and two of five males died before examination on the day following dosing. Abnormal clinical observations in the remaining three males, which died later that day, included severe weakness, vasodilatation, and prostration.

At the 1250 mg/kg dose level, three of five females died on the day following administration of the test material and by Day 2 of the study, an additional female died. The remaining female died before clinical examinations on Day 3 of the study. Abnormal clinical signs on the day of dosing included moderate weakness which progressed to severe weakness, and vasodilatation in all animals. On the day following administration of the test material, only slight weakness was noted in males. In the two surviving females, abnormal clinical signs included severe weakness (2/2), prostration (1/2), and brown urine (1/2). By Day 2 of the study, all males appeared clinically normal and an additional female had died. Abnormal clinical signs noted in the remaining female included severe weakness, brown urine, decreased feces, and hypothermia. This female died the following day. No other abnormal clinical signs were noted, all males survived to scheduled necropsy, and all gained weight normally.

Abnormal clinical signs at the 625 mg/kg dose level were restricted to slight weakness which progressed to moderate weakness, and vasodilitation in all animals on the day of dosing. Brown urine was also noted from one female. On the day following administration of the test material, all animals appeared clinically normal. All animals at this dose level gained weight normally and survived the 14-day observation period.

ACUTE ORAL TOXICITY IN THE RAT CONT.

REMARKS CONT: In the 2500 mg/kg dose group, treatment-related changes observed at necropsy included edema of the glandular gastric mucosa (5/5 males, 5/5 females), congestion of the gastric serosa (4/5 males, 5/5 females), and excessive mucus accumulation in the lumen of the duodenum (5/5 males, 5/5 females).

At the 1250 mg/kg dose level, no treatment-related changes were noted at necropsy of the males. Treatment-related changes in females included edema of the glandular (4/5) and non-glandular (2/5) gastric mucosa, congestion of the gastric serosa (2/5), excessive mucus accumulation in lumen of the duodenum (3/5), and brown discoloration of the inguinal hair (1/5).

No treatment-related changes were observed at necropsy in males or females at the 625 mg/kg dose level.

All other lesions observed at necropsy were not considered treatment-related. No tissue was collected for microscopic examination.

The test article was a gastrointestinal irritant as evidenced by edema of the gastric mucosa, congestion of the gastric serosa, and excessive accumulation of mucus in lumen of the duodenum. The cause of death for rats dying after exposure to the test material was not determined.

ACUTE DERMAL TOXICITY IN RATS

Dose levels tested: 2, 6, and 20 mL/kg

No. Rats/dose: 5 of each sex/dose

Vehicle: None

Preparation: Administered as received

Initial Body weight range (g): 2 and 6 mL/kg = (M) 176 - 196 (F) 160 - 170

20 mL/kg = (M) 271 - 296 (F) 196 - 205

Strain: Crl:CD®(SD)BR

Method of calculation: Weil method

Test SOP No.: TA 310

DOSE mL/kg	NO. RATS DOSED(M,F)	NO. DEATHS (M,F)	TIME OF DEATH	WEIGHT GAIN* 1 WEEK(M,F)	WEIGHT GAIN* 2 WEEK(M,F)
2	5,5	0,0	----------	5+,5+	5+,5+
6	5,5	5,5	Day 1 - Day 2	--,--	--,--
20	5,5	5,5	Day 1	--,--	--,--

* + = Number of animals gaining weight
- = Number of animals losing weight

SUMMARY OF CLINICAL SIGNS		
DOSE (mL/kg)	CLINICAL SIGN	NO. OF RATS AFFECTED
2	Slight Weakness	5M,5F
	Signs at the Application Site	
	- Slight Erythema	5M,5F
	- Necrosis	5M,5F
	- Escars	5M,5F
	- Scarring	5M,4F
	Clinically Recovered	5M,3F
6	Slight Weakness	5M,5F
	Death (Day 1-before clinical observations)	2M,5F
	Severe Weakness	3M
	Prostration	3M
	Signs at the Application Site	
	- Slight Erythema	3M
	- Necrosis	3M
	Death (Day 1-after clinical observations)	1M
	Death (Day 2)	2M

(Continued)

ACUTE DERMAL TOXICITY IN RATS CONT.

SUMMARY OF CLINICAL SIGNS CONT.		
DOSE (mL/kg)	CLINICAL SIGN	NO. OF RATS AFFECTED
20	Slight Weakness Progressing to Severe Weakness	5M,5F
	Prostration	5M,5F
	Vasodilatation	5M,5F
	Death (Day 1)	5M,5F

ESTIMATED DERMAL LD_{50} IN RATS
MALES: 3.6 mL/kg - 95% C.I. 2.2 - 5.6 mL/kg
FEMALES: 3.6 mL/kg - 95% C.I. 2.2 - 5.6 mL/kg

SKIN ABSORPTION: Evident
(Based on Dermal Toxicity study)

REMARKS: The test article was moderately toxic by the dermal route. All doses were applied to the skin after the hair had been removed with an electric clipper. An occlusive wrap was used to hold the test material against the skin for 24 hours, and at the end of exposure, residual test material was washed off with running water.

At the 20 mL/kg dose level, slight weakness was observed in all animals one hour after the start of exposure. By four hours, all animals had clinical signs of severe weakness, prostration, and vasodilatation. All animals died before termination of the 24-hour exposure period.

For the selection of additional dose levels, a range finding study was conducted at dose levels of 2.5, 5, and 10 mL/kg. One male rat was used at each dose level, with body weights ranging between 183-206 grams. Only the animal at the 2.5 mL/kg dose level survived a 7-day period. Based on the range finding studies, additional dose levels of 2 and 6 mL/kg were selected for the dermal toxicity study.

At the 6 mL/kg dose level, slight weakness was noted in all animals during exposure to the test material, and two of five males and all females died before termination of the exposure period. Systemic toxicity observed in the remaining three males that survived the exposure period included severe weakness and prostration. Slight erythema and necrosis were observed at the site of application of the test material. One of three males

ACUTE DERMAL TOXICITY IN RATS CONT.

REMARKS CONT.: died one hour and thirty minutes after termination of the exposure period, and by the following day, the last two males had died.

At the 2 mL/kg dose level, all animals appeared normal during the exposure period. At the end of the exposure period, slight erythema and necrosis at the application site and slight weakness were noted for all animals. Necrosis was observed only at the end of exposure, while erythema persisted to Day 3 of the study. By Day 2 of the study, eschars at the application site were observed on all animals, and by Day 9, scarring was also noted at the application site on nine of ten animals. By termination of the 14-day observation period, animals had either clinically recovered or abnormal signs were limited to escars and scarring. All animals survived to scheduled necropsy, and all gained weight normally.

Treatment-related changes observed at necropsy consisted of focal necrosis of the skin of the back for all animals in the 20 mL/kg dose group, and for four of five males and all females in the 6 mL/kg dose group.

At the 2 mL/kg dose level, treatment-related changes observed at necropsy included an eschar (1/5 females) and scars (2/5 females) on the skin of the back. No treatment-related changes were observed in males.

All other lesions observed at necropsy were not considered treatment-related and no tissue was collected for microscopic evaluation. The cause of death for rats dying after exposure to the test material was not determined.

The test article was a skin irritant as evidenced by necrosis, eschars, and scars on the skin of the back. Percutaneous absorption was evident based on clinical signs of weakness, vasodilatation, and prostration during the exposure period.

ACUTE TOXICITY - DERMAL IRRITATION IN GUINEA PIGS

24 hr. Occluded Single Dose

Slight Moderate Strong Severe

Dose levels tested: 0.5 mL

No. guinea pigs/dose: 5

Vehicle: None

Preparation: Administered as received

Initial Body weight range (g): 414 - 443

Strain: Crl:(HA)BR Hartley

Test SOP No.: TA 160

Sex: Male

Acute Skin Irritation
Slight
Moderate
Strong

Estimated Corrosivity
No
Questionable
Yes

RESULTS				
ANIMAL NO.	MAXIMUM EFFECT		WEIGHT CHANGE (g)	
	SKIN - 24/48 HOURS	SKIN - 2 WEEKS	1 WEEK	2 WEEKS
606	Mod. Ery., Slt. Ed., Mod. N., & Slt. Esc.	Slt. Esc.	+44	+94
607	Mod. Ery., Mod. Ed., Mod. N., & Mod. Esc.	Slt. Esc.	+48	+110
608	Mod. Ery., Slt. Ed., Mod. N., & Slt. Esc.	Slt. Esc.	+45	+94
609	Mod. Ery., Slt. Ed., Mod. N., & Mod. Esc.	Normal	+50	+85
610	Mod. Ery., Mod. Ed., Mod. N., & Mod. Esc.	Slt. Esc.	+40	+75

KEY: Slt. = Slight, Mod. = Moderate, Str. = Strong; Ery. = Erythema, Ed. = Edema, N. = Necrosis; Esc. = Eschar, Scr. = Scarring, V. Slt = Very Slight; St. = Staining, Ero. = Erosion

REMARKS: The test article was a strong skin irritant. A dose of 0.5 mL was applied to each depilated guinea pig abdomen, and an occlusive wrap was used to hold the test material against the skin for 24 hours. During the first 48 hours after removal of the occlusive wrap,

ACUTE TOXICITY - DERMAL IRRITATION IN GUINEA PIGS CONT.

REMARKS CONT: signs of irritation included moderate erythema (5/5), slight (3/5) to moderate (2/5) edema, moderate necrosis (5/5), and slight (2/5) to moderate (3/5) eschar formation at the site of application of the test material. By 72 hours, the two animals with slight eschar formation developed moderate eschars. On Day 7, signs of irritation included slight erythema (1/5) and slight (1/5) to moderate (4/5) eschars. By Day 14, a single animal appeared normal, and slight eschars at the application site persisted in the remaining four animals. All animals survived the 14-day observation period, and all gained weight normally. Based on the absence of clinical signs of toxicity or reduced weight gain, percutaneous absorption was not evident under the conditions of this study.

ACUTE TOXICITY - REPEATED SKIN

Dose levels tested: 0.5 mL drop-on

No. of doses: 9 No. of days: 11

No. guinea pigs/dose: 5

Vehicle: None

Preparation: Administered as received

Initial body weight range (g): 401 - 432

End of study body weight range (g): 429 - 496

Strain: Crl:(HA)BR Hartley

Test SOP No.: TA 170

Sex: Not Determined

RESULTS	GREATEST EFFECT				
	ERYTHEMA	EDEMA	NECROSIS	ESCHAR	OTHER
1ST DOSE	0	0	0	0	0
LAST DOSE	2	2	2	3	0

REMARKS: There was exacerbation of the irritant response with repeated application of the test material. The test material was applied topically, as received, to the backs of guinea pigs after the hair had been removed with an electric clipper. After a single dose, no signs of irritation were noted. At termination of the first week of the study, signs of irritation were limited to slight erythema at the site of application on all animals. After the seventh application over a period of nine days, signs of irritation included erythema, edema, necrosis, and eschar formation. After nine daily applications over a period of eleven days, signs of irritation consisted of moderate erythema (5/5), moderate edema (5/5), moderate necrosis (5/5), and moderate (2/5) to strong (3/5) eschars. Based on the absence of clinical signs of toxicity or reduced weight gain, percutaneous absorption was not evident under the conditions of this study.

ACUTE TOXICITY - SKIN SENSITIZATION
KODAK FOOTPAD METHOD

No. guinea pigs per dose: 10

Irritation drop-on body weight range (g): 417 - 471

Induction body weight range (g): 345 - 471

Strain: Crl:(HA)BR Hartley

Test SOP No.: TA 180

Sex: Male

Estimated
Human Risk
Low
Moderate
High

Irritation Drop-on

Concentrations tested: 0.05 mL of compound in 5.0 mL of Acetone + Dioxane + Guinea Pig Fat.

Highest average score: 0

Induction and Challenge Study

Induction preparation: 0.05 mL of a 1.0% solution of compound in Freund's Complete Adjuvant.

No. of challenge doses: 1

Challenge preparation: 1.0 mL of compound in 10.0 mL of Acetone + Dioxane + Guinea Pig Fat.

SENSITIZATION	NUMBER POSITIVE				TOTAL SCORE
	NORMAL	SLIGHT	MODERATE	STRONG	
FREUND'S ONLY	10				0
1% CPD. IN FREUND'S	10				0

REMARKS: The test article has a low potential to cause human sensitization. No reaction was observed at challenge in any of the animals previously induced with Freund's adjuvant or the test article in Freund's adjuvant.

ACUTE TOXICITY - RABBIT EYE IRRITATION

Dose level tested: 0.1 mL

No. of doses: 1

No. rabbits/dose: 3 washed / 3 unwashed

Vehicle: None

Washing agent: Distilled water

Body weight range: Not determined

Strain: New Zealand white

Test SOP No.: TA 150

Sex: Not Determined

Acute Eye
Irritation
Slight
Moderate
Strong

RESULTS	3 UNWASHED EYES					
	IMMED.	1 HR.	24 HR.	48 HR.	72 HR.	DAY 7
Initial	Slt.					
Conjunctiva		Mod.	Mod.	Mod.	Mod.	
Lids		Mod.	Mod.	Mod.	Slt.	
Nict. Membrane		Mod.	Mod.	Mod.	Mod.	
Corneal Opacity			Mod.			
Iris				Mod.		
Adnexal Stain			2/3			
Corneal Stain			3/3			
Discharge		Mod.				
Number Normal	0	0	0	0	0	3

RESULTS	3 WASHED EYES					
	IMMED.	1 HR.	24 HR.	48 HR.	72 HR.	DAY 7
Initial	Slt.					
Conjunctiva		Mod.	Slt.	Slt.		
Lids		Slt.				
Nict. Membrane		Mod.	Slt.	Slt.		
Corneal Opacity						
Iris						
Adnexal Stain			0/3			
Corneal Stain			0/3			
Discharge		Slt.				
Number Normal	1	0	0	0	3	3

KEY: Slt. = Slight Mod. = Moderate Str. = Strong

ACUTE TOXICITY - RABBIT EYE IRRITATION CONT.

NO. RESPONDING	Slight	Moderate	Strong	Severe	Fluorescein Stain	
					Adnexa	Cornea
UNWASHED EYES		3/3			2/3	3/3
WASHED EYES	3/3				0/3	0/3

REMARKS: The test article was a moderate eye irritant. In unwashed eyes, signs of irritation consisted of moderate erythema and edema of the lids, conjunctivae, and nictitating membranes in all eyes at the one-hour observation period. In addition, moderate discharges were noted from all unwashed eyes one hour after dosing. Slight corneal opacity was evident in unwashed eyes only at the 24-hour observation period. When unwashed eyes were tested with fluorescein dye 24 hours after dosing, there was corneal staining in all eyes and staining of the nictitating membranes in two of three eyes. Injection of the iris was noted in each unwashed eye at the 48-hour observation period. By Day 7 of the study, all unwashed eyes appeared clinically normal.

Immediate washing was palliative. In washed eyes, signs of irritation included moderate erythema (3/3) and slight edema (2/3) of the conjunctivae and nictitating membranes and slight erythema of the lids (3/3) at the one-hour observation period. A slight discharge from all washed eyes was noted one hour after dosing. No corneal opacity was noted in washed eyes at any time during the 7-day observation period. At 24 hours, no corneal or adnexal staining were observed when washed eyes were tested with fluorescein dye. By 72 hours, all washed eyes appeared normal.

Q.A. INSPECTION STATEMENT
(CFR 58.35(B)(7) 792.35(B)(7) 160.35(B)(7))

STUDY: 89-0081-1 STUDY DIRECTOR: TOPPING, D.C.
ACCESSION NUMBER: 907670

STUDY TYPE: ACUTE TOXICITY REPORT

M. L. James
(AUDITOR, QUALITY ASSURANCE UNIT)

1/25/90
DATE

THIS STUDY WAS INSPECTED BY 1 OR MORE PERSONS OF THE QUALITY ASSURANCE UNIT OF THE HAEL, EASTMAN KODAK COMPANY, ROCHESTER, N.Y. AND WRITTEN STATUS REPORTS WERE SUBMITTED ON THE FOLLOWING DATES:

INSPECTION DATES	STUDY/PHASE INSPECTED	STATUS REPORT DATES
07/24/89	ACUTE DERMAL IRRITATION TEST PROTOCOL APPENDIX SUBMISSION	
07/24/89	REPEATED SKIN IRRITATION PROTOCOL APPENDIX SUBMISSION	
07/26/89	SENSITIZATION PROTOCOL APPENDIX SUBMISSION IRRITATION DROP-ON	
07/31/89	RABBIT EYE IRRITATION PROTOCOL APPENDIX SUBMISSION	
08/01/89	ACUTE DERMAL TOXICITY (LD50) PROTOCOL APPENDIX SUBMISSION DOSE CALCULATIONS TEST ARTICLE APPLICATION TO TEST SYSTEM	
08/02/89	SENSITIZATION PROTOCOL APPENDIX SUBMISSION FOOT PAD INJECTION	
08/02/89	ACUTE ORAL LD50 PROTOCOL APPENDIX SUBMISSION	
08/07/89	ACUTE ORAL LD50 NECROPSY	
08/08/89	ACUTE DERMAL TOXICITY (LD50) PROTOCOL APPENDIX SUBMISSION TEST ARTICLE WEIGHT DOSE CALCULATIONS TEST ARTICLE APPLICATION TO TEST SYSTEM	
08/09/89	SENSITIZATION 1 WEEK CHALLENGE (TEST ARTICLE-CARRIER-MIXTURE DROP-ON)	08/09/89
01/25/90	ACUTE TOXICITY REPORT FINAL REPORT REVIEW	01/25/90